# Anaerobic fermentation

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# 1. Materials and methods

## 1.1. Batch fermentation

The conversion of biomass to biogas was tested according to the widely accepted German standard protocol DIN 1734. Completely sealed, 0.5 liter laboratory fermenters were used for these experiments. The glass flasks were filled with the biomass to be degraded and the headspaces were replaced with ultrapure nitrogen gas by repeated vacuum and gassing cycles in order to establish anaerobic environment. The anaerobic degradation (AD) fermentation was initiated using a mixed and enriched methanogenic community of microbes, which originated from a large scale AD biogas plant. The "batch" fermenters were kept at 37°C using a laboratory incubator with intermittent shaking. This is the typical and standard arrangement for a „batch" AD fermentation. It is important to note that in these experiments the biomass degradation and conversion to biogas is carried out in a closed system. Biogas production is a result of microbiological activity in the closed vessel as long as there is biomass available for fermentation. Thereafter biogas production gradually decreases as the biodegradable biomass in the fermenter is consumed. This experimental condition is therefore strikingly different from the technologies employed in industrial scale AD fermentation, where the substrate addition and effluent removal is done continuously or intermittently. Consequently, "batch" biogas tests carried out according to the DIN 11734 protocol are suitable for the determination of the theoretical biogas potential of the substrate rather than to measure or simulate the actual biogas yield in a continuous or semi-continuous AD fermenter.

The biogas produced in the "batch" fermenters is collected in a second glass vessel, which is filled up with water and connected to the AD reaction vessel using silicone rubber tubing. The evolved gas displaces an equal volume of water into a third vessel, still in a closed system, and the volume of the dislocated water can be measured daily. The principle of the arrangement is presented in Figure 1. The biogas productivity values are corrected to normal atmospheric conditions and calculated according to the organic material input.

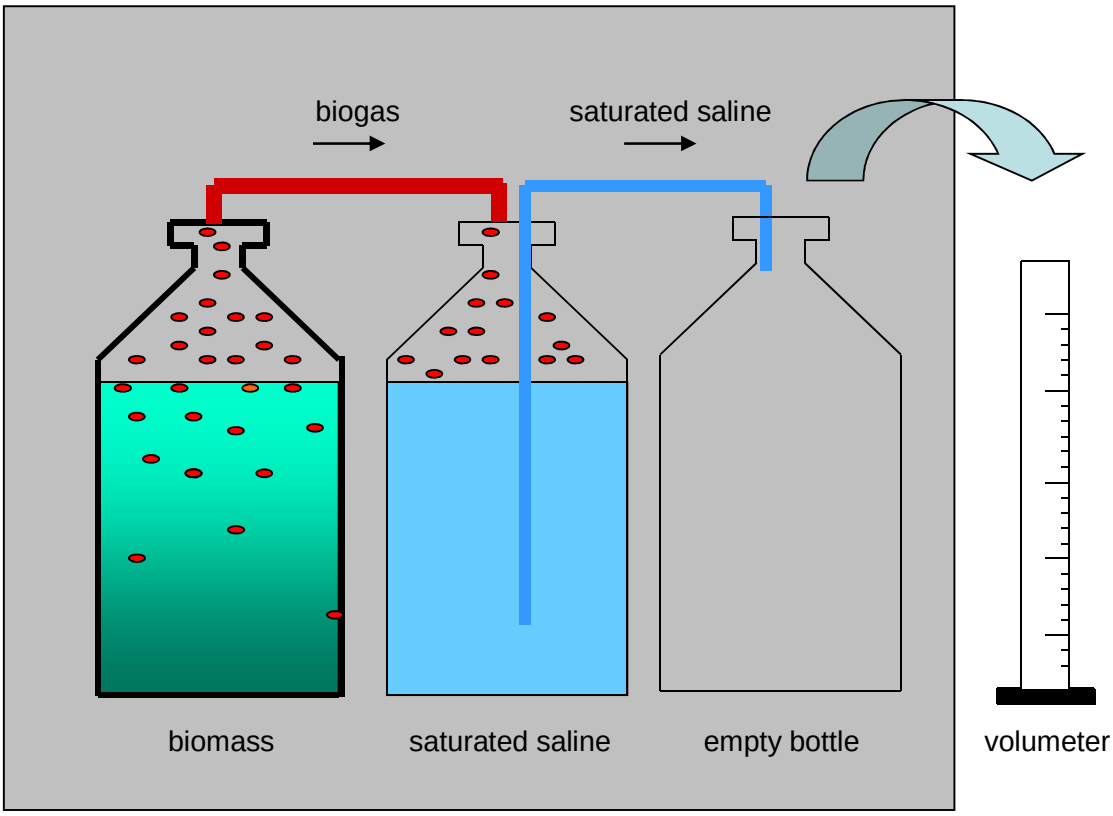


Figure 1. Experimental set-up.

### 1.2. Gas analysis

Composition of the evolved biogas was measured by taking 100 µl aliquots from the headspace and injecting the gas mixture into a gas chromatograph (Shimadzu GC-2010) equipped with a Carboxen 1010 column and a TCD detector. Ultrapure nitrogen was used as carrier gas.

### 1.4. Dry matter and organic dry matter content determination

The dry matter content was quantified by drying the biomass at 105 ºC overnight and weighing the residue. Further heating of this residue at 550 ºC until its weight did not change yielded the organic dry matter value.

### 1.5. Substrate biomass

The substrate biomass was ferentation residue and recrement. The TS and the oTS values of the samples are the following:

| | fermentation residue | recrement |
|---|---|---|
| TS(%) | 8,2 | 47,4 |
| oTS(%/TS) | 70,2 | 87,3 |

Figure 2.: Substrate biomass TS and oTS values

## 2. Results

The results of the batch tests are found on figure 3.

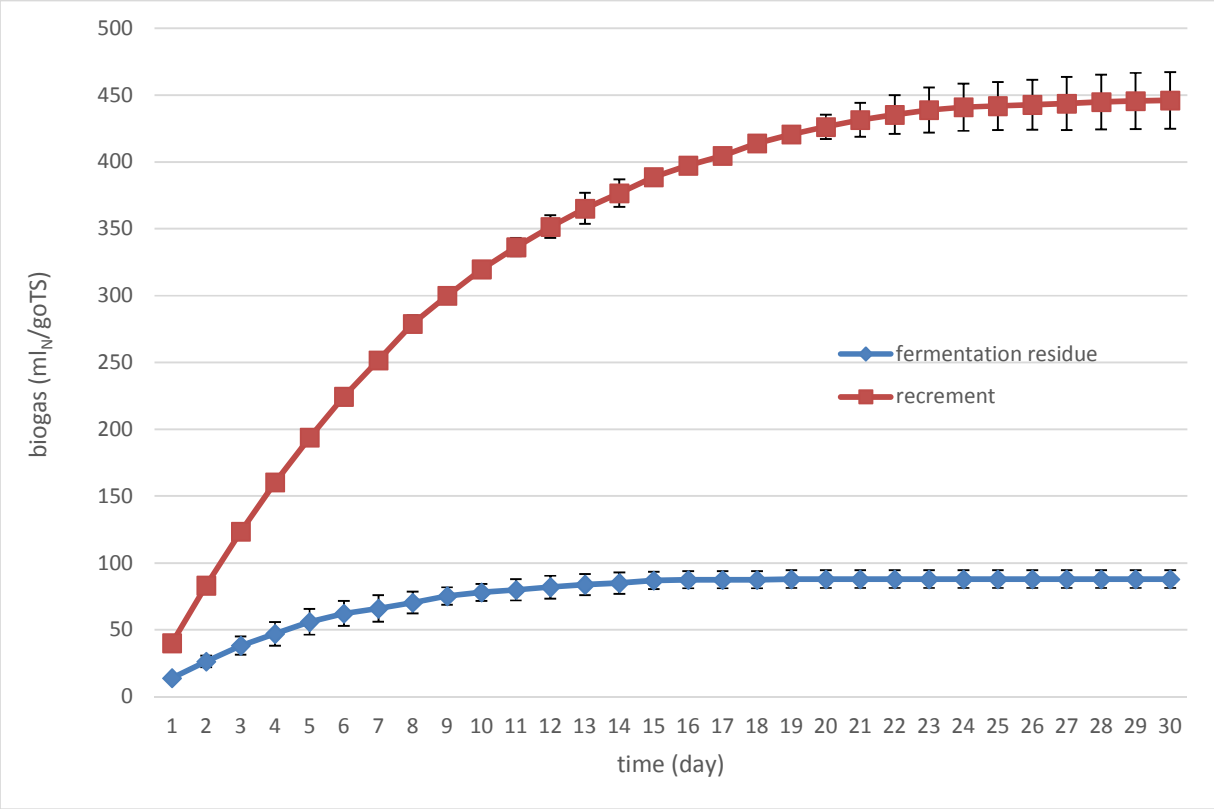


**Figure 3: Biogas production**

The biogas yield of the substrates are found in the next table:

| | fermentation residue | recrement |
|---|---|---|
| biogas yield (ml$_N$/g oTS) | 87,9 | 446,1 |
| biogas yield (m$^3$/t wet weight) | 5,06 | 184,6 |
| methane content (%) | 56,9 | 57,4 |